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Early Effects of Catecholamine Therapy on Mucosal Integrity, Intestinal Blood Flow, and Oxygen Metabolism in Porcine Endotoxin Shock

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Objective

To determine the early effects of therapy of endotoxin (ET) shock with epinephrine, norepinephrine, or dopexamine on splanchnic circulation, oxygen metabolism, sigmoid mucosal pHi, bacterial translocation, and morphologic integrity of the ileal, colonic, and sigmoid mucosa.

Summary Background Data

Conflicting concepts exist concerning the catecholamine therapy of septic shock, but little is known about the effects of catecholamine treatment on splanchnic circulation and mucosal integrity.

Methods

ET shock was induced in pigs by ET infusion over 30 minutes, and animals were studied for 4 hours. All animals were resuscitated with fluid. To mimic the treatment of septic shock in humans, mean arterial pressure was maintained in two groups at >70 mm Hg with the administration of epinephrine or norepinephrine. A third group of animals received dopexamine at 7 μg/kg per minute.

Systemic and splanchnic blood flow and oxygen metabolism were studied, sigmoid colon mucosal pHi was obtained tonometrically, and bacterial translocation was determined by culture of portal venous blood, mesenteric lymph nodes, liver, spleen, and lung specimens. Histologic sections of ileal, colonic, and sigmoid mucosa were morphometrically examined for therapy effects.

Results

All investigated catecholamines increased cardiac output and systemic oxygen delivery, whereas intestinal blood flow and oxygen delivery remained unchanged. Sigmoid mucosal pHi decreased in all study animals, but the decrease was most pronounced in the epinephrine group. Pigs receiving epinephrine also showed >40% damage of the mucosa of the ileum and colon, whereas animals receiving ET alone, norepinephrine, or dopexamine showed only moderate lesions with signs of restitution. No animal showed bacterial translocation.

Conclusions

Systemic hemodynamics and oxygen metabolism data do not reflect intestinal perfusion. Norepinephrine or dopexamine administration in ET shock causes no additional impairment of intestinal integrity. Epinephrine therapy, in contrast, is associated with a significant reduction of mucosal pHi and considerable early mucosal damage. Its application in septic shock is hazardous.

Septic shock still represents a major cause of death in intensive care patients.[1,2] It is associated with hypotension, low cardiac output, and subsequent inadequate tissue perfusion, potentially leading to end organ failure. In the intestine, this impaired perfusion may lead to endothelial hyperpermeability, subsequent leakage of microorganisms or endotoxin (ET) to lymphatic or blood vessels, and a resulting enhancement of the septic state. The gut as a physiologic reservoir of bacteria has therefore been regarded as the "motor" of multiple organ failure during sepsis.[3] Increased intestinal mucosal permeability for viable pathogens has been well defined[4,5] and has been observed as a consequence of conditions such as endotoxemia,[6,7] local

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acidosis,[8] intestinal hypoperfusion,[9,10] or thermal injury,[11,12] all of which are associated with compromised intestinal blood flow.

Therapeutic concepts of septic shock aim at an elevation of cardiac output by generous fluid resuscitation and administration of inotropic catecholamines. A decrease in systemic vascular resistance, which leads to reduced perfusion pressure in vital organs, is counterbalanced by the administration of vasopressors.[13] This symptomatic therapy of hypotension and hypoperfusion represents generally accepted treatment.[13]

Although these means have been shown to improve systemic hemodynamics, their effects on regional perfusion are not well defined. The choice of catecholamines is controversial.[14] For example, epinephrine has been repeatedly recommended because of its positive inotropic and vasopressive properties.[15–17] Other authors favor the use of norepinephrine in facultative combination with beta-mimetic substances.[18–21] Further, several studies indicate that vasoactive drugs influence intestinal perfusion differently.[16,18,21,22] Breslow et al.[23] studied the effects of various catecholamines on organ blood flow in a porcine model of ET shock and found that vasopressors had no effects on intestinal blood flow. In contrast, others showed a marked reduction of intestinal blood flow as a result of norepinephrine administration.[24] Conflicting results were also reported from studies in humans, which suggested a detrimental effect of certain vasopressive agents on intestinal pHi.[21]

The synthetic beta-mimetic catecholamine dopexamine has been attributed to have a protective effect in patients with sepsis,[25] although its value in patients with septic shock is not fully clarified.[26]

A recent review on the effects of catecholamines on regional perfusion emphasizes the scarcity of data on this topic.[22]

The exact impact of catecholamine treatment of septic shock on intestinal integrity is not known. Therefore, this study compared the effects of various regimens of ET shock treatment on systemic and intestinal hemodynamic and oxygen metabolism parameters in the pig. We also investigated sigmoidal mucosal pHi, the morphology of ileal, colonic, and sigmoid mucosa obtained from these animals, and bacterial translocation to portal venous blood, mesenteric lymph nodes, liver, spleen, or lung at the end of *in vivo* experiments.

MATERIALS AND METHODS

The study protocol was approved by the animal research committee of the medical faculty of the University of Vienna, and complied with the animal research and animal care guidelines of the Austrian Ministry of Science.

Animal Preparation

Twenty-three landrace pigs weighing 16 to 22 kg each were housed at the Center for Surgical Research of the University of Vienna. Animals were fasted for 24 hours before the experiments, with free access to water. The pigs were anesthetized with intramuscular ketamine (20 mg/kg) and pentobarbital sodium (2 mg/kg). After cannulation of an ear vein, maintenance of analgoanesthesia was established with piritramide (1.5 mg/kg per hour). Relaxation was established with pancuronium bromide (0.02 mg/kg per hour). Pigs were placed in the supine position on a heating blanket (38° to 39°C) and orotracheally intubated. Mechanical ventilation was provided with 40% oxygen-enriched air by a constant-volume time-cycled ventilator (Servo 900C, Siemens–Elema, Stockholm, Sweden). Tidal volume (8 to 12 ml/kg), ventilatory rate (10 to 15 cycles per minute), and FiO_2 were adjusted to keep PO_2 >90 mm Hg and pCO_2 between 35 and 44 mm Hg at the end of the equilibration period and thereafter remained unchanged. The initial infusion rate for all animals was 25 ml/kg per hour of Ringer's solution. The infusion rate for experimental animals was adapted after the start of the experiment, as described below. Before surgery, a silicone balloon catheter (TRIP Sigmoid Catheter, Tonometrics Inc., Worcester, MA) was transanally introduced into the sigmoid colon for tonometric pHi measurement. The correct position of the catheter was confirmed by palpation after laparotomy.

Surgical Procedure

All surgical procedures were performed under strictly sterile conditions. Through a left lateral neck incision, the carotid artery was dissected and cannulated with a catheter for measurement of mean arterial pressure (MAP) and withdrawal of blood samples. The left internal jugular vein was cannulated for fluid and drug administration. Through the left subclavian vein, a balloon-tipped pulmonary artery thermodilution catheter (P575-15CM-EH, 5.5F, Abbott Laboratories, Chicago, IL) was inserted and advanced into correct position according to typical pulmonary artery flow curves. Using this catheter, right atrial pressure, pulmonary artery pressure, and pulmonary capillary wedge pressure (PCWP) were measured. Cardiac output was determined by the thermodilution method and was recorded by a cardiac output computer (Oximetrix 3 So2/CO Computer, Abbott Laboratories) in triplicate and averaged.

Blood gas analysis was performed on a blood gas analyzer (AVL 995-Hb, AVL, Graz, Austria) and on a cooximeter (OSM 2 Hemoxidation, Radiometer Copenhagen, Copenhagen, Denmark).

After the abdomen was opened with a midline incision, ileocecal mesenteric lymph nodes and biopsy samples of the liver were obtained. Thereafter, a suprapubic puncture catheter was inserted into the bladder and secured with a pursestring ligature. The portal vein and the superior mesenteric artery were gently dissected, and electromagnetic flow probes (Type B, Hellige, Freiburg, Germany) in adequate sizes were attached to these vessels. A 5F silicone catheter was inserted into the portal vein downstream of the attached

Table 1. MEAN AND MAXIMAL DOSE OF CATECHOLAMINES (μG/KG/MIN) TO MAINTAIN MEAN ARTERIAL PRESSURE BETWEEN 70 AND 80 MM HG AND INFUSION RATS OF RINGERS SOLUTION (ML/KG/HR) TO MAINTAIN PCWP AT PREENDOTOXIN LEVELS

Group	C	FR	FR/E	FR/NE	FR/DX
Mean catecholamine	–	–	1.1 ± 0.2	0.96 ± 0.2	7.0 ± 0
Max. catecholamine	–	–	2.4 ± 0.6	2.1 ± 0.5	7.0 ± 0
Mean infusion rate	28 ± 3	36 ± 2.5	34 ± 2	36 ± 2	42 ± 3*
Max. infusion rate	28 ± 3	45 ± 7	46 ± 4	46 ± 8	65 ± 2†

* $p < 0.05$ FR/DX *vs.* FR/E.
† $p < 0.05$ FR/DX *vs.* all other groups.
PCWP = pulmonary capillary wedge pressure; C = controls; FR = fluid resuscitation; FR/E = fluid resuscitation plus epinephrine hydrochloride; FR/NE = fluid resuscitation plus norepinephrine hydrochloride; FR/DX = fluid resuscitation plus dopexamine hydrochloride.

flow probe for pressure measurements and blood sampling. Flows and pressures were documented online and recorded on a multichannel recorder (Hellige, Freiburg, Germany). Thereafter, the abdomen was closed, and the animals were allowed to stabilize for 30 minutes.

Experimental Protocol

At time 0, animals were continuously infused with either 250 μg/kg ET (*Escherichia coli* O111B4, Difco Laboratories, Detroit, MI) in 50 ml 0.9% saline solution (study animals, n = 19) or vehicle (50 ml 0.9% saline solution) alone (controls, n = 4) for 30 minutes. They were subsequently monitored for 240 minutes.

Study animals were randomly divided into four groups. The first group (n = 4) received fluid resuscitation (FR) alone. After the ET infusion, FR (Ringer's solution) was increased from 25 ml/kg per hour to an individual amount to maintain PCWP at the pre-ET level. The second group (n = 5) received FR as in the first group, plus epinephrine hydrochloride (FR/E) individually administered to maintain a MAP of 70 to 80 mm Hg. The third group (n = 5) received FR as in the first group, plus norepinephrine hydrochloride (FR/NE) individually administered to maintain a MAP of 70 to 80 mm Hg. The fourth group (n = 5) received FR as in the first group, plus dopexamine hydrochloride (FR/DX), administered at the maximum recommended dosage of 7 μg/kg per minute.

To mimic the therapy of septic shock in humans, catecholamine treatment of groups 2 through 4 was not started until a decline of MAP was observed. After that, the drugs were administered to achieve the defined target level of MAP.[23]

The mean and maximum catecholamine dosages, as well as mean and maximum amounts of fluid administration, for the study animals are shown in Table 1.

All hemodynamic parameters and mucosal pHi were recorded at minutes 0, 30, 60, 120, 180, and 240. Blood samples were taken from the carotid artery, pulmonary artery, and portal vein at the same points of time for blood gas analysis. Ten milliliters of portal venous blood was drawn into special tubes (Wampole Isolator 10, Merck, Darmstadt, Germany) for microbiologic analysis at cannulation of the portal vein as well as at 240 minutes. After the completion of 240 minutes, the abdomen was reopened under sterile conditions, and biopsy samples of the mesenteric lymph nodes, liver, and spleen were harvested. A lung biopsy sample was obtained through an incision in the diaphragm. Finally, specimens of the gut were obtained as described below. Feces were obtained to determine the bacteria present in the gut. At the end of the experiments, the animals were killed by a bolus injection of saturated potassium chloride solution.

Histology and Morphometry

Five-centimeter segments of the distal ileum, ascending colon, and sigmoid colon were removed and opened longitudinally. External muscle layers were removed by blunt dissection, and mucosal preparations were prepared for light microscopy as previously described.[27] Mucosal damage was assessed by a histopathologist (RS), who performed morphometry on coded, paraffin-embedded, serial vertical sections stained with hematoxylin and eosin, as previously described.[27–29] Approximately 3 cm of mucosa was morphometrized for every specimen. We used a Leitz Diaplan research microscope (Wild Leitz Ltd., Heerbrugg, Switzerland, with objective magnification ×4, ×6.3, ×16, ×25, and ×40 and ocular magnification ×12.5), a Panasonic color CCD video camera (model WV-CD 130/G, Matshushita, Japan), an analogue/digital monitor screen (model PVM-1271Q, superfine pitch, Sony, Japan), and a personal computer (IBM PS/2, model PS2; IBM, Armonk, NY), extended by insertion of a frame grabber (ITI PC Visionplus board; Imaging Technology Corp., Woburn, MA) and a graphic tablet (Summasketch plus 12″ × 12″ MM 1201; Summagraphics Corp., Fairfield, CT). The extent of damage was measured in micrometers and given as a percentage of total mucosal surface. The system was calibrated using a micrometer slide (2 mm in 200 equal parts; Wild Heerbrugg, Switzerland). Histologic criteria for epithelial damage were increased eosinophilic staining intensity of epithelial cells, karyopyknosis, karyolysis, and karyorrhexis, cell disruption, formation of subepithelial blebs, and lifting off of cells from the basal lamina.[27]

Microbiology

The specimens of mesenteric lymph nodes, liver, spleen, and lung were weighed and placed in grinding tubes (Aigner, Vienna, Austria) containing isotonic NaCl (1 ml/100 mg material). The microbiologic analysis was performed qualitatively and quantitatively from the homogenized specimens on Koch blood agar, Columbia agar, and MacConkey and Schaedler plates. Incubation was done under both aerobic and anaerobic conditions at 37°C for 72 hours with inspections performed every 24 hours. Identification of the bacteria was performed by the API system (Analytab Products, Bio Merieux, France). The results were documented as colony-forming units (CFU) per gram of material.

Wampole tubes with the specimens of portal venous blood were centrifuged in a 35° fixed-angle centrifuge for 30 minutes at 3000 rounds per minute; the supernatant was withdrawn and discarded, and the remaining sediment was vortexed for 10 seconds. The microbiologic analysis was again performed qualitatively and quantitatively on Koch blood agar, Columbia agar, and Schaedler plates, with incubation and identification as for the organ specimens. The results were documented as CFU per milliliter of blood.

Statistical Analysis

The time course of hemodynamic variables and pHi values was analyzed using the multivariate approach to analysis of variance for repeated measures using the base value at time 0, the five therapy groups, the four time points (60, 120, 180, and 240 minutes), and the interaction between group and time as independent explaining factors. Including the base value as an independent covariate corrects for differences that may exist at time 0. The interaction between groups and time is significant if at least one group behaves differently from other groups over time. Contrasts were calculated for specific comparisons when group-by-time interactions were significant. Adjustments for multiple comparisons were made according to Tukey-Kramer. Results of morphometry were analyzed by analysis of variance. Data are expressed as mean ± SEM. Observed significance levels <0.05 were regarded as statistically significant.

RESULTS

Systemic and Splanchnic Hemodynamic Parameters

Control animals showed an unaffected course of all hemodynamic parameters up to 240 minutes of observation. In

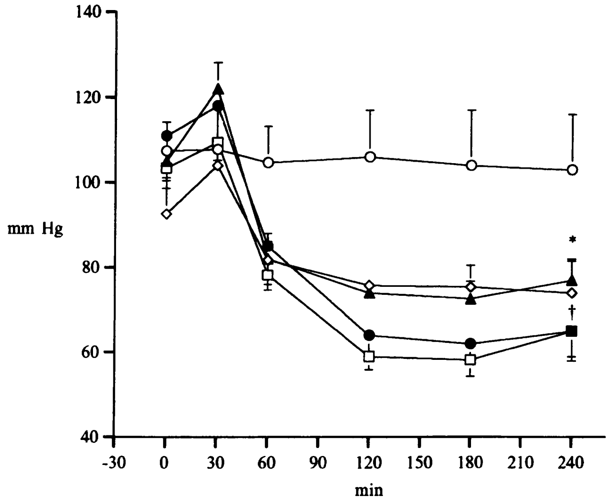

Figure 1. Time course of MAP. From t = 0 to t = 30, study animals received 250 μg/kg ET as a continuous infusion. Individual catecholamine therapy started shortly thereafter (see Table 1). Open circles, controls; closed circles, FR; open rhombus, FR/E; closed triangle, FR/NE; open square, FR/DX. *p < 0.01 for groups FR/E and FR/NE over time *vs.* controls; †p < 0.01 for groups FR and FR/DX over time *vs.* controls.

study animals, mean pulmonary artery pressure uniformly increased from 18 ± 0.7 mm Hg at time 0 to 49 ± 1.6 mm Hg at 30 minutes. Cardiac output correspondingly decreased during the same period from 2.5 ± 0.1 liters to 1.8 ± 0.2 liters. MAP intermittently increased to 112 ± 3 mm Hg at the end of the ET infusion and then began to decrease below baseline levels at 35 ± 3 (FR/E), 34 ± 2 (FR/NE), and 37 ± 2 (FR/DX) minutes. At these times, catecholamine therapy was induced.

With FR alone, MAP continuously decreased to 62 ± 5 mm Hg at 180 minutes and then slightly recovered to 65 ± 6 mm Hg at 240 minutes (p < 0.01). Vasopressor therapy maintained blood pressure from time 60 at 76 ± 5 mm Hg (FR/NE) and 77 ± 5 mm Hg (FR/E), whereas administration of FR/DX did not stabilize blood pressure (p < 0.01 controls *vs.* all other groups over time) (Fig. 1).

Although MAP decreased in all study animals during the observation period, cardiac index (ml/kg per minute) increased at 240 minutes in groups FR/E (208 ± 30), FR/NE (187 ± 11), and FR/DX (207 ± 17) and decreased in the FR group (35 ± 9; p < 0.01 for group differences and p < 0.02 for group-time interaction) (Fig. 2). Absolute flow of the superior mesenteric artery (ml/minute) decreased in all groups, although no statistically significant group differences were observed (Fig. 3). PCWP did not change significantly during the experiment in any of the groups.

Systemic and Splanchnic Oxygen Metabolism

According to changes in cardiac index, the systemic oxygen delivery index (ml/minute) at 240 minutes signifi-

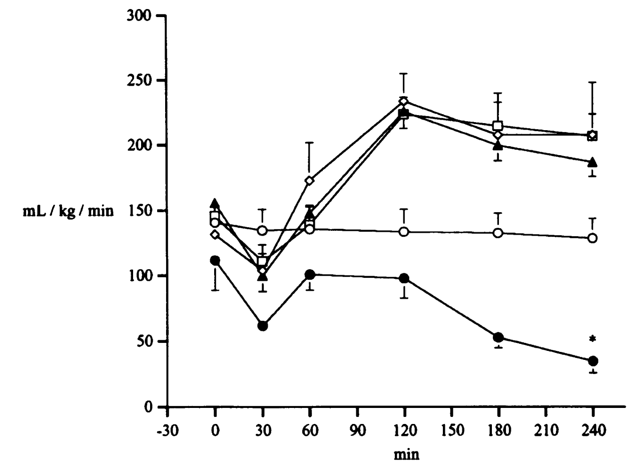

Figure 2. Time course of cardiac index. From t = 0 to t = 30, study animals received 250 μg/kg ET as a continuous infusion. Individual catecholamine therapy started shortly thereafter. Open circles, controls; closed circles, FR; open rhombus, FR/E; closed triangle, FR/NE; open square, FR/DX. $p < 0.02$ for groups FR, FR/E, FR/NE, and FR/DX over time *vs.* controls. *$p < 0.01$ for FR over time *vs.* controls, FR/E, FR/NE, and FR/DX.

cantly increased in group FR/E (2.7 ± 0.5), FR/NE (2.5 ± 0.2), and FR/DX (2.6 ± 0.2), remained unchanged in controls (1.6 ± 0.02), and decreased significantly in group FR (0.79 ± 0.19) ($p < 0.001$ for group-time interaction) (Table 2). The systemic oxygen extraction index change was not significantly different for groups over time. Intestinal oxygen delivery followed intestinal blood flow without significant changes. Splanchnic oxygen consumption and extraction rates did not differ significantly (Table 3).

Intestinal Mucosa pHi

As shown in Figure 4, tonometrically assessed pHi of the sigmoid colon significantly decreased in all experimental groups over time when compared with the pHi of controls ($p < 0.001$ for group-time interaction). This decrease was most pronounced in the FR/E group and reached statistical significance when compared with groups FR/NE and FR/DX ($p < 0.001$).

Histology and Morphometry

Light microscopy examinations of controls revealed excellent preservation of normal ileal, colonic, and sigmoid mucosal architecture. In contrast, histology of the FR, FR/NE, and FR/DX groups showed moderate damage of ileal, colonic, and sigmoid surface epithelial cells. The colonic epithelium was focally damaged. Although the lesions were seen in the upper compartment of the mucosa, the crypts remained intact. Epithelial cells were shrunken and rounded up, and most of the nuclei were pyknotic or presented karyorrhexis. Several damaged cells were detached from the lumen (Figs. 5 and 6). The ileum showed a similar focal damage pattern: villi were shortened by approximately one third (Figs. 7 and 8). In the FR group, signs of restitution, with flattened epithelial cells, were detected. In the FR/E group, histology showed severe damage of ileal, colonic, and sigmoid crypt and surface epithelium. In these instances, the mucosa was generally damaged, including all epithelial compartments (Figs. 9 and 10). In none of the mucosal preparations were signs of inflammation observed in the lamina propria.

Morphologic damage, expressed as the percentage of damaged mucosa, was dependent on therapy and segment (Fig. 11). Again, the FR, FR/NE, and FR/DX groups showed minor, not statistically significant damage of the ileal and colonic mucosa. The sigmoid mucosa showed significant damage in FR animals. FR/E animals had moderate damage to the sigmoid mucosa and considerable damage to the ileal and colonic mucosa (see Fig. 11).

Translocation of Viable Bacteria

In all animals, cultures of portal venous blood, liver, spleen, and lung showed no growth of bacteria. In two animals in the control group and one animal in the FR group, 25 and 50 CFU of *E. coli*, respectively, were detected in a mesenteric lymph node harvested at the beginning of the experiment but not in the mesenteric lymph node retrieved at the end of the experiment.

DISCUSSION

The intestine, because it is a potential source of bacteria in patients with sepsis and septic shock, has attracted considerable attention in the past years. Although bacterial

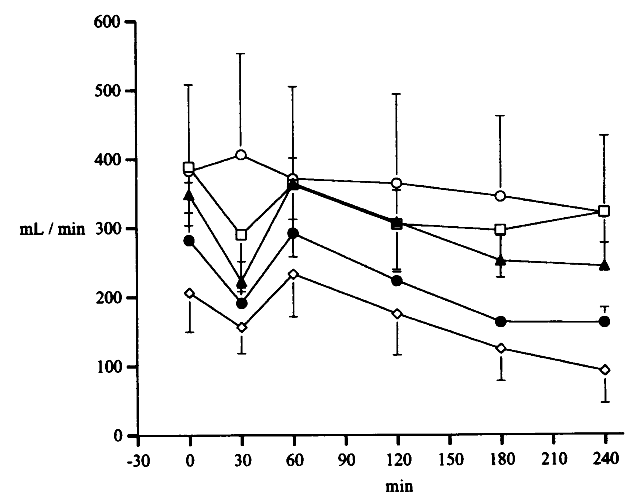

Figure 3. Time course of superior mesenteric artery flow. From t = 0 to t = 30, study animals received 250 μg/kg ET as a continuous infusion. Individual catecholamine therapy started shortly thereafter (see Table 1). Open circles, controls; closed circles, FR; open rhombus, FR/E; closed triangle, FR/NE; open square, FR/DX. p = NS for time-by-group interaction.

Table 2. SYSTEMIC OXYGEN METABOLISM DATA

Variable/Group	Time (min) 0	30	60	120	180	240
DO2i ml/kg/min						
C	16 ± 1	18 ± 1	18 ± 1	17 ± 1	17 ± 0	16 ± 1
FR	17 ± 2	12 ± 1	17 ± 2	17 ± 2	11 ± 2	8 ± 2
FR/E	17 ± 3	13 ± 1	23 ± 3	31 ± 3	28 ± 4	27 ± 5*
FR/NE	22 ± 2	16 ± 2	23 ± 1	30 ± 2	26 ± 2	25 ± 2*
FR/DX	19 ± 2	16 ± 2	20 ± 2	29 ± 2	28 ± 3	26 ± 2*
VO2i ml/kg/min						
C	6 ± 0.5	6 ± 0.4	6 ± 0.6	7 ± 0.6	6 ± 0.3	6 ± 0.8
FR	5 ± 0.3	5 ± 0.5	5 ± 0.3	6 ± 1.0	5 ± 0.9	3 ± 0.2
FR/E	6 ± 1.0	6 ± 0.5	7 ± 2.0	9 ± 2.3	9 ± 2.4	8 ± 2.6
FR/NE	6 ± 0.6	7 ± 0.8	8 ± 0.5	8 ± 0.6	6 ± 1.5	7 ± 0.4
FR/DX	6 ± 0.1	6 ± 0.4	6 ± 0.7	7 ± 0.4	7 ± 0.7	8 ± 1.0
O2ER (%)						
C	33 ± 4	32 ± 3	34 ± 2	39 ± 1	37 ± 2	38 ± 3
FR	33 ± 4	37 ± 5	31 ± 2	33 ± 4	45 ± 6	47 ± 9
FR/E	34 ± 7	46 ± 4	30 ± 6	31 ± 6	31 ± 4	33 ± 2
FR/NE	29 ± 1	49 ± 9	34 ± 1	26 ± 1	23 ± 6	31 ± 2
FR/DX	31 ± 2	40 ± 3	33 ± 2	25 ± 1	25 ± 1	31 ± 5

* $p < 0.002$ for time effect per group (60–240 min).
Data presented as means ± SEM.
DO2i (systemic oxygen delivery index) is calculated as: cardiac output/bodyweight × arterial oxygen content VO2i (systemic oxygen consumption index) is calculated as: cardiac output/bodyweight × (arterial oxygen content-mixed venous oxygen content) O2ER (systemic oxygen extraction ratio) is calculated as: VO2/DO2.
C = controls; FR = fluid resuscitation; FR/E = fluid resuscitation plus epinephrine hydrochloride; FR/NE = fluid resuscitation plus norepinephrine hydrochloride; FR/DX = fluid resuscitation plus dopexamine hydrochloride.

translocation in these conditions has been shown and thoroughly investigated,[5] there is little knowledge about the consequences of conventional treatment of septic shock with different catecholamines on intestinal integrity or permeability.[22]

Established therapeutic measures of cardiocirculatory support—in particular vasopressor[13] and inotropic therapy[30,31]—have been scrutinized recently.[14,32,33] Together with new methods for the monitoring of intestinal perfusion,[34,35] new interest in the splanchnic circulation of critically ill patients has evolved.[36] Several observations indicating a distinct influence of different vasoactive drugs on intestinal perfusion have been reported.[21,22,33] We therefore sought to investigate the different effects of various catecholamines on pathophysiologic, morphologic, and microbiologic parameters during ET shock in an established, well-defined animal model.[23,37,38]

In applying conventional catecholamine treatment for septic shock, we were able to improve systemic blood pressure or to achieve the desired increase in cardiac output.[20,30] The doses of catecholamines we used match those reported for the treatment of human septic shock.[15,18,20] Fluid resuscitation was administered in accordance with other studies that used a similar setting,[23,37] and infusion rates resulted in fluid administration of up to 1200 ml/hour; this is equivalent to approximately 4500 ml/hour when converted to a human body weight of 70 kg. Despite that, intestinal macrocirculation remained unchanged or deteriorated. This effect might in part be attributed to an autoregulatory effect of the intestinal macrocirculation, which has been described as the "autoregulatory escape" after continuous stimulation of regional sympathetic nerves; this mechanism allows the intestinal circulation to gain partial or total recovery of flow.[39]

Both systemic and intestinal oxygen delivery followed closely the course of the hemodynamic data, resulting in no improvement in intestinal oxygen supply despite the achievement of a considerable increase in systemic oxygen delivery. Oxygen extraction and consumption did not change significantly over time, and the choice of catecholamine did not result in significant differences. In accordance with other authors,[23] we could not demonstrate significant differences in overall intestinal blood flow with different catecholamines in the treatment of ET shock, but we found signs of distinct distribution of the delivered blood flow by tonometry and histology.

During recent years, determination of gastric or sigmoid mucosal pHi has been added to the diagnostic armamentarium of intensive care medicine as a means to determine malperfusion of the intestinal mucosa. It has been shown to correlate with outcome in different patient cohorts.[34,35] In a similar porcine model, intestinal pHi was reduced by lipopolysaccharide infusion[37] and the tonometrically assessed hydrogen ion concentration compared well with pHi as assessed by intraepithelial electrodes.[37] The control group in our experiments exactly mirrors the findings of these

Table 3. INTESTINAL OXYGEN METABOLISM DATA

	Time (min)					
Variable/Group	**0**	**30**	**60**	**120**	**180**	**240**
DsO2 ml/min						
C	42 ± 2	43 ± 3	39 ± 2	38 ± 4	37 ± 4	36 ± 3
FR	43 ± 6	33 ± 3	49 ± 4	37 ± 3	28 ± 2	27 ± 4
FR/E	28 ± 7	21 ± 5	32 ± 8	23 ± 8	17 ± 6	13 ± 7
FR/NE	49 ± 3	35 ± 3	55 ± 4	41 ± 6	33 ± 5	33 ± 5
FR/DX	52 ± 13	43 ± 11	53 ± 16	41 ± 10	39 ± 10	42 ± 12
VsO2 ml/min						
C	9 ± 2	10 ± 3	9 ± 3	8 ± 4	10 ± 6	10 ± 3
FR	9 ± 2	10 ± 2	12 ± 2	14 ± 3	14 ± 2	12 ± 3
FR/E	11 ± 4	11 ± 4	14 ± 5	11 ± 4	9 ± 3	7 ± 3
FR/NE	16 ± 1	19 ± 3	19 ± 3	16 ± 2	13 ± 2	13 ± 2
FR/DX	16 ± 3	15 ± 4	13 ± 2	13 ± 2	12 ± 3	11 ± 2
O2sER (%)						
C	21 ± 5	24 ± 5	23 ± 4	21 ± 3	25 ± 6	26 ± 5
FR	22 ± 6	34 ± 9	24 ± 5	37 ± 7	47 ± 7	48 ± 10
FR/E	36 ± 6	43 ± 7	41 ± 5	41 ± 5	55 ± 7	61 ± 9
FR/NE	34 ± 3	57 ± 8	35 ± 4	39 ± 2	41 ± 3	43 ± 3
FR/DX	33 ± 5	35 ± 3	31 ± 5	36 ± 5	36 ± 5	30 ± 2

p = not significant.

DsO2 (splanchnic oxygen delivery) is calculated as: superior mesenteric artery flow × arterial oxygen content VsO2 (splanchnic oxygen consumption index) is calculated as: portal venous flow × (arterial oxygen content-portal venous oxygen content) O2sER (splanchnic oxygen extraction ratio) is calculated as: VsO2/DsO2.

C = controls; FR = fluid resuscitation; FR/E = fluid resuscitation plus epinephrine hydrochloride; FR/NE = fluid resuscitation plus norepinephrine hydrochloride; FR/DX = fluid resuscitation plus dopexamine hydrochloride.

authors. However, vasopressor therapy of ET shock obviously cannot improve this condition and may even aggravate the mucosal acidosis. Dopexamine, however, maintained pHi during endotoxemia, probably a result of the well-preserved mucosal blood flow during ET shock. Because overall superior mesenteric artery blood flow did not differ significantly among the groups, differences in pHi can most likely be attributed to a different partition of blood flow to the layers of the bowel wall.[39] Our results are in agreement with the results of recent clinical studies describing distinct effects of different catecholamines on intestinal

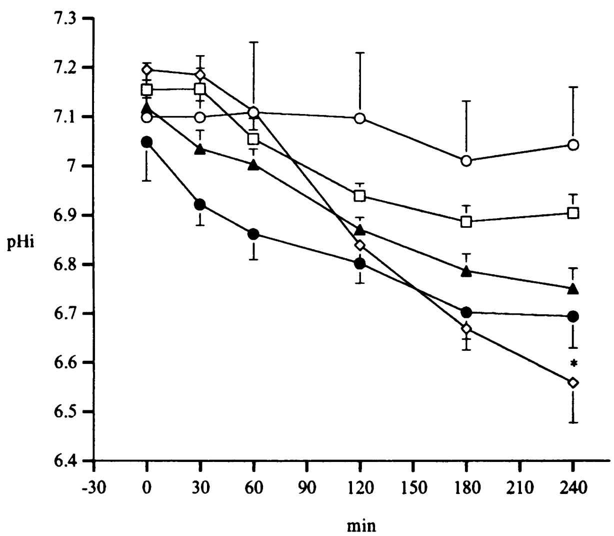

Figure 4. Time course of sigmoid mucosal pH. From t = 0 to t = 30, study animals received 250 μg/kg ET as a continuous infusion. Individual catecholamine therapy started shortly thereafter (see Table 1). Open circles, controls; closed circles, FR; open rhombus, FR/E; closed triangle, FR/NE; open square, FR/DX. $p < 0.01$ for FR, FR/E, FR/NE, and FR/DX over time *vs.* controls. *$p < 0.01$ for FR/E over time *vs.* controls, FR/DX, and FR/NE.

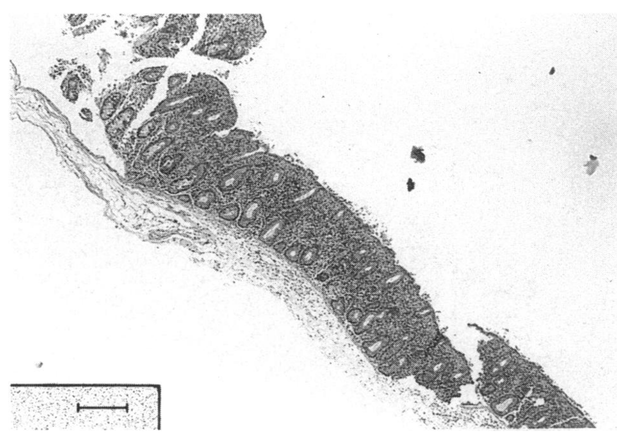

Figure 5. Histologic section of colonic mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR animal showing focal damage. Lesions are present in the upper compartment of the mucosa; crypts remain intact. Epithelial cells are shrunken and rounded up, and many of the nuclei are pyknotic or present karyorrhexis. Several damaged cells are detached from the lumen.

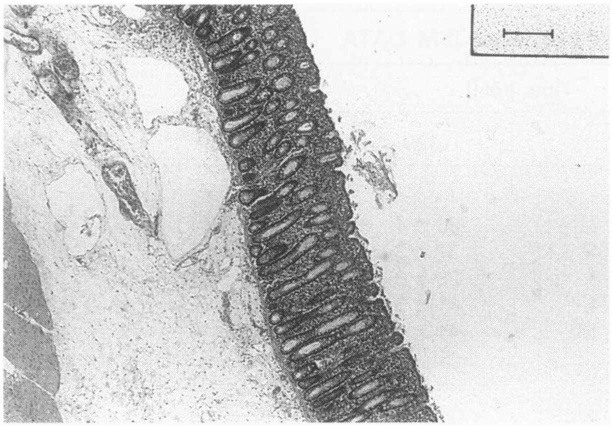

Figure 6. Histologic section of colonic mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR/NE animal showing focal damage. Present damage is similar to that in FR animals, with lesions in the upper compartment of the mucosa (shrunken epithelial cells, karyorrhexis), whereas crypts are intact. Also in this specimen, damaged cells are detached from the lumen.

Figure 8. Histologic section of ileal mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR/NE animal, again showing focal damage pattern, as was present in mucosal preparations from FR animals.

mucosal pHi.[21,22] In patients with hyperdynamic sepsis, studied for a similar observation period of 3 hours, norepinephrine was found to be superior to high-dose dopamine with respect to gastric mucosal pHi preservation.[21]

In addition to mucosal pHi data, our morphologic findings provide further evidence of contrasting effects of different vasopressors and inotropes on morphologic integrity, despite similar effects on systemic hemodynamics. Results of mucosal morphometry are in agreement with tonometric data. We were able to show that FR/NE and FR/DX administration caused only minor damage of the ileal and colonic epithelium (see Fig. 9), and FR/NE and FR/DX reduced ET-induced endothelial defects in the sigmoid. In contrast, FR/E therapy after only 3 hours caused significant mucosal damage in all investigated regions of the gut. Further, mucosal preparations of animals receiving ET alone or in combination with FR/NE or FR/DX showed clear signs of restitution of epithelial discontinuities, whereas this was not observed with epinephrine-treated animals.

The fact that despite the observed mucosal damage, viable bacteria did not translocate through the intestinal barrier remains difficult to explain. It may be attributed to the short period of observation. This finding is in accordance with other researchers, who demonstrated that neither ischemia nor ischemia–reperfusion of the gut in a porcine model was followed by translocation of bacteria to blood and tissue specimen after 8.5 hours.[40] Tokyay et al.[38] demonstrated the single and additive effects of relevant trauma on bacterial translocation in the pig. In their study, anesthesia, surgery, and ET administration led to no or only insignificant translocation, whereas burn in combination with ET administration or resuscitation caused significant translocation of via-

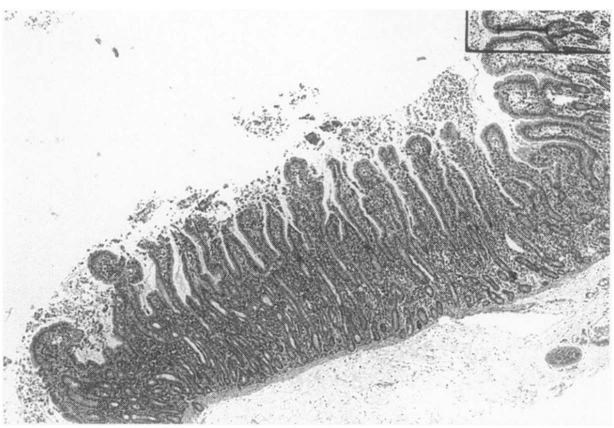

Figure 7. Histologic section of ileal mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR animal showing focal damage. Histologic signs of mucosal damage similar to the colonic mucosa with shrunken epithelial cells, pyknotic or karyorrhectic nuclei, and damaged cells detached from the lumen. Villi are shortened by approximately one third.

Figure 9. Histologic section of ileal mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR/E animal showing severe generalized damage of all epithelial compartments.

ble bacteria at 48 hours after trauma. Translocation in the pig model therefore seems to be induced less easily than in rodent models.

On the other hand, killing of transmigrated bacteria by the natural host defense mechanisms of study animals may have occurred, thus avoiding the spill of viable pathogens to the lymphatics and the portal vein. Nevertheless, killing of gram-negative bacteria may still cause the release of ET, thereby inducing the inflammatory cytokine cascade.[41] Kane et al.[42] showed the presence of fragments of killed bacteria using sensitive methods in blood specimens from burned mice. However, killing of bacteria in the mucosa or submucosa should lead to some sign of inflammation, and we found no histologic evidence of inflammation or bacterial fragments in any of the investigated mucosa specimens.

In conclusion, hyperdynamic systemic hemodynamics induced by catecholamine therapy have little effect on the intestinal macrocirculation; however, different drugs may cause distinct disturbances of the microcirculation. With regard to intestinal integrity, epinephrine does not seem to be a suitable therapeutic option in septic shock: its administration led to signs of severely impaired intestinal microcirculation and considerable mucosal damage early in treatment. Severe alterations of the mucosa could lead to increased permeability for intestinal pathogens with a longer course of therapy. Norepinephrine, in contrast, seems to provide effective circulatory support, allowing better maintenance of intestinal microcirculation and epithelial morphology. Although dopexamine alone cannot improve blood pressure, it purportedly has positive effects on the splanchnic circulation in septic shock,[25] a finding that correlates well with the preservation of intestinal pHi and intestinal integrity demonstrated here. As to maintenance of intestinal barrier function, these two substances, perhaps given in combination, seem to be a favorable therapeutic option to achieve the desired hemodynamic conditions in the treatment of septic shock.

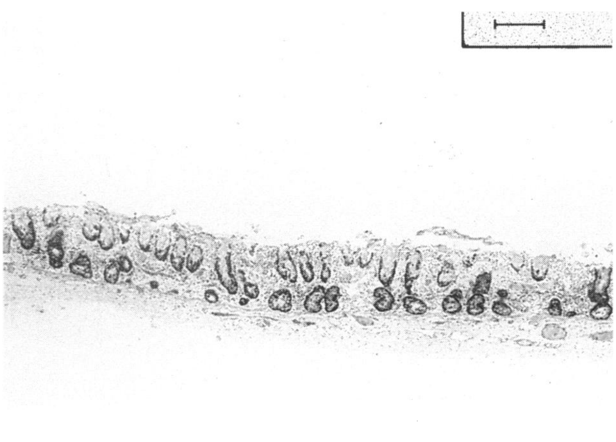

Figure 10. Histologic section of colonic mucosa, hematoxylin and eosin staining, ×50 magnification; internal scale represents 200 μm. Specimen from an FR/E animal showing the same magnitude of damage of all epithelial compartments as in the ileum of FR/E animals.

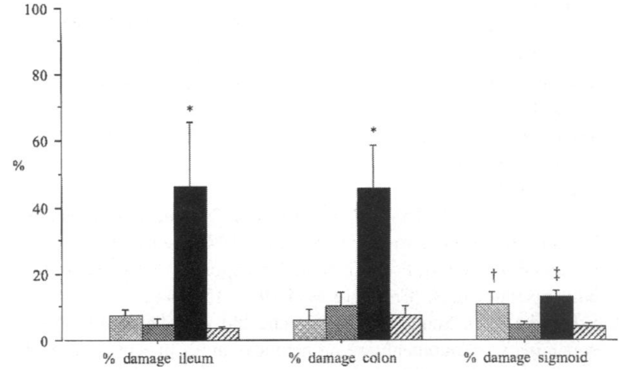

Figure 11. Percentage damage of ileal, colonic, and sigmoid mucosa. Intact epithelium (no damage) for controls. Light-gray columns, FR; dark-gray columns, FR/DX; black columns, FR/E; striped columns, FR/NE. *$p < 0.05$ for FR/E *vs.* all other groups, †$p < 0.02$ for FR *vs.* controls, ‡ $p < 0.02$ for FR/E *vs.* controls, FR/NE, and FR/DX.

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